REMARKS

Claims 1-28, 31, 44, 50, 58-60 and 65-67 are canceled without prejudice, claims 1-13, 27 and 28 being previously withdrawn. Applicants reserve the right to pursue any of the canceled subject matter in one or more continuing applications. Claims 29, 32, 41, 45 and 61-64 have been amended. No new matter has been added. Claims 29, 30, 32-43, 45-49, 51-57, 61-64 and 68-73 are pending and under examination.

The present amendment overcomes the Examiner's rejections and places the application in condition for allowance. Therefore, in accordance with MPEP §714.12, Applicants respectfully request that the present amendment be entered and that the application proceed to allowance.

The Claims

The claims are directed to a method of obtaining pancreatic islet cells. The method includes: (a) providing differentiated or adult pancreatic cells substantially free of islet cells, (b) allowing the differentiated or adult pancreatic cells to proliferate to form a population of dedifferentiated pancreatic cells, (c) adding a component of extracellular matrix (ECM) to the population of dedifferentiated pancreatic cells; and (d) growing the cells in the presence of ECM for a time sufficient for the cells to express insulin.

The claims have been amended to specifically recite the steps of allowing the adult differentiated or adult cells to proliferate to form dedifferentiated cells. Dedifferentiated cells have lost their differentiated phenotype and have reverted to a pluripotent state. The claims also specifically include the step of growing the cells in the presence of ECM for a time sufficient for the cells to express insulin.

The amendments are supported by the original claims and throughout the application, e.g., page 2, lines 1-5; page 5, lines 1-8; page 32, lines 5-7.

Rejections Under 35 U.S.C. § 102

Claims 14, 15, 21-26 and 65-67 are rejected as anticipated by Kerr Conte [IDS-AH]. These claims have been canceled without prejudice, thereby obviating the rejection. Accordingly, the rejection should be withdrawn.

Rejections Under 35 U.S.C. § 103

Gmyr

Claims 14, 21-26, 29, 32, 36-42, 45, 47-51 and 65-73 are rejected as unpatentable over Gmyr et al. [IDS-AG] (Gmyr). The Examiner argues that method "C" of Gmyr discloses "the similar, if not identical, protocol" as the claims because:

> the result of the cited method, the obtaining of pancreatic islet cells is an intrinsic result in the method "C" as disclosed by Gmyr et al. The same starting cells, cultured in the same manner, can be reasonably expected to produce the same product.
>
> ***
>
> [t]he protocol as disclosed by Gmyr comprises step of obtaining dedifferentiated cells and step of culturing dedifferentiated cells in the presence of ECM. Thus, it results in the pancreatic islet cells within the meaning the claims [sic]. The structural differences in the methods for obtaining islet cells which are intended by applicants are uncertain as claimed and as argued.

Claims 14, 15, 21-26 and 65-67 are canceled. The rejection has been met, in part, by amending the remaining claims for clarity and to specifically recite an affirmative step of allowing the adult differentiated or adult cells to proliferate to form dedifferentiated cells (rather than defining dedifferentiated cells as a product by process limitation). The claims have also been amended to recite growing the dedifferentiated cells in the presence of ECM for a time sufficient for the cells to express insulin. These steps are neither disclosed nor suggested in Gmyr (this is also discussed in more detail below). The rejection is traversed in part, in particular with regard to the Examiner's interpretation of the teachings of Gmyr.

As discussed in the enclosed declaration under 37 C.F.R. §1.132 of Dr. Bonner-Weir (hereinafter "the Bonner-Weir declaration") Gmyr relates to expansion of ductal cells from various types of tissue. Gmyr does not disclose or suggest that insulin-producing islet cells (as

required by the claims) are produced by any of methods A, B or C. Gmyr merely states "We are now screening various conditions to induce the endocrine differentiation of these ductal precursor cells" (emphasis added). This is clearly no more than an invitation to experiment. Gmyr does not even speculate about what such conditions might be.

As discussed in the Bonner-Weir declaration, the term "dedifferentiated cells" as used by Gmyr means something completely different than the term as used in the present specification and claims. "Dedifferentiated cells" as defined in the specification and recited in the claims, are adult or differentiated pancreatic cells, substantially free of islet cells, that have been allowed to proliferate, have lost their differentiated phenotype and are pluripotent. (See specification at page 2, lines 1-5; page 17, lines 13-15; page 32, line 6 et seq.) The present claims recite a step of allowing the adult or differentiated cells to proliferate. In contrast, as used by Gmyr, "dedifferentiated cells" are ductal cells that have arisen from a population of exocrine cells. Gmyr describes method C as follows:

> exocrine cells [i.e., acinar cells] were allowed to dedifferentiate to a ductal phenotype in culture. (. . .) the cell yield appeared virtually unlimited (. . .) Immunohistochemical studies confirmed the ductal epithelial (cytokeratin 19, CA 19-9) nature of the cells. (. . .) [Method C] allowed further expansion of ductal epithelial cells when cultured in 804G matrix with HGF or EGF. Conclusion: (. . .) exocrine tissue dedifferentiation in culture allowed the obtention of sufficient numbers of human ductal pancreatic cells and their further in-vitro expansion. We are now screening various conditions to induce the endocrine differentiation of these ductal precursor cells. (Emphasis added.)

Thus, in what Gmyr calls "method C", Gmyr describes the expansion of ductal cells from exocrine tissue. Gmyr refers to the expansion of ductal cells from exocrine tissue in culture as "dedifferentiation." As discussed in the enclosed declaration under 37 C.F.R. §1.132 of Dr. Bonner-Weir (hereinafter "the Bonner-Weir declaration"), **"dedifferentiation" as used by Gmyr, does not include proliferation**. In fact, the *in vitro* expansion of ductal cells from exocrine (acinar) tissue (which is the phenomenon disclosed by Gmyr) has been shown by others to not involve cell division. See, for example, Rooman et al. (2000) *Diabetes* 43:907-914 (enclosed with the Bonner-Weir declaration). Although Rooman was published after the

relevant filing date, it discusses references describing the obtention of ductal cells from cultured exocrine pancreas prior to the filing date. (See Rooman, page 912, first sentence of Discussion) Rooman concludes as follows:

> a nine fold increase in cells with ductal characteristics [from acinar exocrine tissue] with 26 to 64% preservation of initial DNA, and **the absence of cell division** as assessed by incorporation of BrdU, excluded the possibility of selective survival or overgrowth by a small contaminating population of centroacinar-ductular cells. (. . .) The present results are indicative of a process termed "direct transdifferentiation", in which **cellular phenotypic conversion is independent from DNA-replication**. (Rooman, page 912, 1st paragraph of Discussion, emphasis added.)

Accordingly, the expansion of exocrine cells into ductal cells, which is the phenomenon disclosed by Gmyr, does not involve cell division, i.e., it does not involve cell proliferation. Rather, the expansion of exocrine cells into ductal cells is a mechanism in which exocrine cells acquire characteristics of ductal cells during culture *in vitro* without passing through a proliferative (cell division) phase, as required by the claims. Indeed, nothing in Gmyr indicates that the exocrine cells were allowed to proliferate, as recited in the claims. Nor does Gmyr suggest that proliferation is required or desirable for the expansion of ductal cells, much less for the production of islet cells from exocrine or ductal tissue, as recited in the claims. Contrary to the Examiner's argument, the expression of cytokeratin 19 (as described in Gmyr) is a marker for ductal epithelial cells. Cytokeratin 19 is not a marker for proliferating cells that have lost their ductal phenotype. This much is clear from Gmyr itself, which states: "Immunohistochemical studies confirmed the ductal epithelial (cytokeratin 19,)CA19-9) nature of the cells in each of the three methods" (emphasis added). See also Bouwens et al. (1995) Identification of rat pancreatic duct cells by their expression of cytokeratins 7, 19 and 20 in vivo and after isolation and culture. *J. Histochem. Cytochem.* 43:245-53 (abstract enclosed). Therefore, the fact that the Gmyr cells express cytokeratin 19 merely confirms that the cells produced by the Gmyr method are ductal cells. Gmyr does not suggest or even speculate that the resulting cells of method "C" express insulin, as required by the present claims. Indeed, since ductal cells do not produce insulin, the Gmyr cells cannot reasonably be expected to be the same as Applicants'

dedifferentiated cells, which Applicants have shown to form islet buds and express insulin upon contacting with a component of ECM.

Applicants note that, although this is a rejection under §103, part of the Examiner's argument appears to be based on the belief that Gmyr inherently anticipates the claimed method. Although the above discussion relates to non-obviousness, it is also clear that Gmyr does not inherently anticipate the claims. Indeed, even if there were a possibility that cells produced from the Gmyr method express insulin, this would not be sufficient for inherent anticipation. In order to inherently anticipate, the prior art must necessarily function in accordance with-or include- the claimed limitation. Inherency may not be established by probabilities or possibilities. The mere fact that a certain thing may result from a given set of circumstances is not sufficient. See *Continental Can v. Monsanto*, 948 F.2d 1264 (Fed. Cir. 1991).

In sum, as amended, the claims require affirmative steps of allowing the adult or differentiated cells to proliferate and growing the dedifferentiated cells in the presence of ECM for a time sufficient for the cells to express insulin. For the reasons discussed above, these steps are not disclosed (either explicitly or inherently) or suggested by Gmyr. Gmyr relates to expansion of ductal cells from exocrine tissue. Gmyr does not provide a motivation or reasonable expectation of success for a skilled artisan to obtain islet cells by any method, much less by the claimed method.

Kerr-Conte or Gmyr taken with US 5,681,587, US 6,077,692, WO 96/40872, Carlsson, and Kerr-Conte [AY]

Claims 14, 15, 17-26, 29, 30, 32-43, 45-57, 61-64 and 66-73 remain rejected as unpatentable over Kerr-Conte or Gmyr taken with US 5,681,587 (US '587), US 6,077,692 (US '692), WO 96/40872, Carlsson, and Kerr-Conte [AY]. The rejection has been met by canceling claims 14, 15, 21-26 and 65-67 and amending the remaining claims. The presently amended claims are limited in that the starting adult or differentiated cells are substantially free of islet cells. All the pending claims include affirmative steps of allowing the adult or differentiated cells to proliferate and culturing the dedifferentiated cells with ECM for a time

sufficient for the cells to express insulin. The present claims are patentable over the cited references for at least the following reasons.

Kerr-Conte describes the expansion of ductal epithelial cells from a preparation of 80% pure adult islet cells that were placed directly on collagen or Matrigel. Kerr-Conte does not disclose or suggest starting with pancreatic cells substantially free of islet cells. If anything, Kerr-Conte teaches away by indicating that one should start with purified islets. Kerr-Conte also does not disclose or suggest the desirability or necessity of allowing the adult cells to proliferate prior to addition of ECM, as recited in the claims. Applicants again note that the ductal epithelial cells described in Kerr-Conte are positive for carbohydrate antigen 19-9, which is a marker of differentiated ductal cells, not of dedifferentiated cells, as discussed above and in the Bonner-Weir declaration. Thus, there is simply no suggestion or motivation in Kerr-Conte to arrive at the claimed methods.

Gmyr also fails to provide the required suggestion or motivation required for a prima facie case of obviousness. The Examiner states that Gmyr "teach[es] the use of extracellular matrix (ECM) in the method of obtaining islet cells..." This is simply not a correct reading of Gmyr. As discussed in detail above, Gmyr does not provide any method to make islet cells from a starting material of exocrine cells in vitro. Gmyr only discloses how to expand exocrine cells into duct cells in vitro. Further, Gmyr does not suggest that allowing adult or differentiated cells to proliferate before contacting with ECM is necessary to obtain duct cells, much less insulin-producing islet cells, from exocrine cells in vitro.

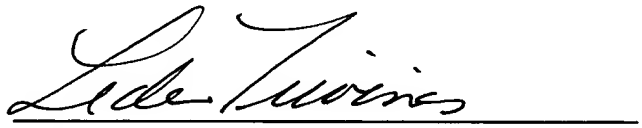
The remaining references do not provide the required teaching or suggestion missing from Kerr-Conte or Gmyr. The remaining references are cited for their disclosure of ECM (US 4,829,000 and US '587), keratinocyte growth factor (US '692), markers of pancreatic cell expansion in vivo (WO 96/40872 and Carlsson), culturing of duct cells on a substrate (US '521), and expansion of duct cells in culture (Kerr-Conte [IDS-AY]). Alone or in combination, none of the other cited references provide the missing disclosure, suggestion or motivation to allow differentiated or adult pancreatic cells substantially free of islet cells to proliferate to form a population of dedifferentiated pancreatic cells, and thereafter add a component of extracellular

matrix (ECM) sufficient for the cells to express insulin. The Examiner has picked a number of secondary references, each of which discloses a limitation recited in one or more dependent claim. However, there is no motivation in any one or in the combination of cited references to perform the specifically recited steps of the claimed methods together. The fact that independent limitations can be found in the prior art does not mean that the invention is obvious unless there is a motivation to combine the steps and limitations into the specifically claimed method. The Examiner has provided no evidence of such a motivation here. A showing of a suggestion, teaching, or motivation to combine "must be clear and particular...Broad conclusory statements regarding the teaching of multiple references, standing alone, are not 'evidence.'" In re Dembiczack, 175 F.3d 994 (Fed. Cir. 1999). Thus, the Examiner's broad, generic basis for finding motivation is insufficient. Accordingly, a prima facie case of obviousness has not been made and Applicants respectfully request that the rejection be withdrawn.

Enclosed is a Petition for Extension of Time along with the required fee. Please apply any other charges or credits to deposit account 06-1050.

Respectfully submitted,

Date: 8 January 2004



Leda Trivinos
Reg. No. 50,635

Fish & Richardson P.C.
225 Franklin Street
Boston, MA 02110-2804
Telephone: (617) 542-5070
Facsimile: (617) 542-8906